

2  
3 METHODS FOR POLYMERIZATION OF ELECTRONIC  
4 AND PHOTONIC POLYMERS

5  
6 CROSS-REFERENCE TO RELATED APPLICATIONS

7 This is a continuation-in-part of U.S. Application Serial No.  
8 09/994,998, filed November 27, 2001, in the names of Sukaut  
9 Tripathy, et al, which, in turn, claims the benefit of U.S.  
10 Provisional Application No. 60/253,109, filed November 27, 2000,  
11 both of which are hereby incorporated herein by reference.

12  
13 STATEMENT OF GOVERNMENT INTEREST

14 The invention described herein may be manufactured and used  
15 by the U.S. Government for governmental purposes without the  
16 payment of any royalty thereon.

17  
18 BACKGROUND OF THE INVENTION

19 Recently, there has been an increased interest in tailored  
20 development of polyaromatic polymers, particularly polyaromatic  
21 polymers that are electrically conductive and/or have useful  
22 optical properties. Examples of electrically conductive polymers  
23 include certain polyanilines, polythiophenes, polypyrroles, and  
24 polyphenols. These conductive polyaromatic polymers may be used  
25 in a variety of electronic devices, including electro-chromic

1 devices, light-emitting diodes, electrostatic discharge  
2 protection, and light weight batteries. Of these polyaromatic  
3 polymers, polyanilines are the most extensively studied, due  
4 largely to superior electrical properties, such as high discharge  
5 capacity.

6 In addition to the above-named electrical properties, thermal  
7 and structural properties of polyphenols have long been exploited.  
8 In particular, phenol-formaldehyde resins, such as novolacs and  
9 resols, have found wide application as wood composites, laminates,  
10 foundry resins, abrasives, friction and molding materials,  
11 coatings and adhesives, fiber binders, and flame retardants. The  
12 use of formaldehyde in polyphenol synthesis, however, presents a  
13 significant toxicological and environmental hazard.

14 Despite the industrial utility of polyaromatic polymers,  
15 their synthesis remains problematic. Known difficulties in the  
16 synthesis of such polymers include inconsistent product  
17 composition, due in part to extensive branching of the polymers.  
18 In addition, many of the polyaromatic polymers are insoluble, or  
19 sparingly soluble, in common solvents, leading to poor  
20 processability. The use of toxic reagents, as noted above, is  
21 another undesirable feature of current synthetic methods. A  
22 search for new methods of synthesizing polyaromatic polymers has  
23 not heretofore yielded a commercially viable approach.

24 Many of the synthetic approaches to forming polyaromatic  
25 polymers use a heme-containing enzyme to catalyze the

1 polymerization. Any such catalyst must necessarily be stable and  
2 active under acidic conditions, as acidic conditions are required  
3 in order to synthesize an electrically conductive form of a  
4 polyaromatic polymer, such as polyaniline.

5 The increasing environmental problem of hazardous chemical  
6 wastes has led to an upsurge in efforts toward the development of  
7 biochemical alternatives for synthesis of electronic and photonic  
8 polymers. Enzymatic polymerizations have attracted much attention  
9 with oxidative enzymes, such as horseradish peroxidase (HRP),  
10 being used for the synthesis of polyanilines and polyphenols  
11 through oxidative free radical coupling reactions.

12 Unfortunately, HRP and other peroxidases are inactive at low  
13 pH and are prohibitively expensive to use commercially. Hematin  
14 has been used to mimic the catalytic activity of HRP. However,  
15 despite its lower costs, hematin is a non-ideal catalyst for  
16 commercial polymerizations because of its low solubility in  
17 acidic, aqueous media. The low solubility of hematin under these  
18 conditions leads to an extremely low rate of polymerization and  
19 very poor yields.

20 The mechanism for HRP catalyzed polymerization involves the  
21 interaction of the heme-iron cofactor of the enzyme with the  
22 peroxide yielding an oxidized heme-iron complex. Subsequently,  
23 the oxidized heme-iron complex reacts with the substrate in a one-  
24 electron transfer reaction to produce the substrate radical and a

1 new iron-heme complex followed by the coupling of the radicals to  
2 form the polymer.

3 This enzymatic approach has not been extended to  
4 polythiophenes or polypyrroles, which have high electrical  
5 conductivity. This is because monomers, such as (3,4)-  
6 ethylenedioxythiophene (EDOT) and pyrrole (PYR), complexed with  
7 the active site of the enzyme catalyst cause deactivation of the  
8 latter and have proved to be unsuitable substrates for this  
9 enzymatic polymerization. This deactivation phenomenon  
10 drastically limits the prospects for the enzymatic synthesis of a  
11 wide range of polymers for possible industrial applications. The  
12 present invention evolved from exploration of the possibility of  
13 usage of a modified hydroxy ferriprotoporphyrin Hematin to serve  
14 as a catalytic center.

15 There is a need for a low cost, high efficiency means of  
16 synthesizing polyaromatic electronic and photonic polymers, which  
17 means is compatible with conditions required to synthesize  
18 polymers with commercially desirable properties.

#### 20 SUMMARY OF THE INVENTION

21 The invention generally is directed to a derivatized hematin,  
22 to a method of forming assembled and derivatized hematins, and to  
23 methods for polymerizing an aromatic monomer with an assembled  
24 hematin or a derivatized hematin.

1           Accordingly, an object of the present invention is to provide  
2           a novel method for the synthesis of a conducting complex such as  
3           poly (3,4)-ethylenedioxythiophene/sulfonate polystyrene  
4           (PEDOT/SPS).

5           A further object of the present invention is to provide a  
6           novel method for the synthesis of a conducting complex of  
7           polypyrrol/sulfonate polystyrene (PPYR/SPS).

8           A further object of the present invention is to provide a  
9           novel method for the synthesis of a conducting complex of  
10          copolymers PPYR-PEDOT/SPS.

11          A further object of the present invention is to provide a  
12          method which results in the production of copolymers Polyaniline-  
13          PPYR/SPS, which have electrical and chemical stability, and  
14          improved processability.

15          A still further object of the present invention is to provide  
16          a method which results in the production of copolymers  
17          Polyaniline-PEDOT/SPS which have electrical and chemical  
18          stability, and improved processability.

19          Still another object of the present invention is to provide a  
20          method which results in the production of copolymers Polyaniline-  
21          PEDOT-PPYR/SPS, which have electrical and chemical stability, and  
22          improved processability.

23          A still further object of the present invention is to provide  
24          a method which results in the synthesis of a Polyaniline-PPYR-  
25          PEDOT/SPS polymer complex wherein the optical and electronic

1 properties of the final complex can be tailored and optimized by  
2 judicious choice or modification of an electrolyte matrix  
3 material.

4 With the above and other objects in view, a feature of the  
5 invention is the provision of hematin derivatized with one or more  
6 non-proteinaceous amphipathic groups, wherein the preferred  
7 amphipathic group is polyethylene glycol, and the hematin  
8 derivatized is soluble over a pH range of about pH 0.5 to about  
9 pH 12.

10 In accordance with a further feature of the invention, there  
11 is provided a method for preparing a derivatized hematin by  
12 reacting hematin with an amphipathic compound. In a preferred  
13 embodiment, the hematin is derivatized with an amphipathic  
14 compound in the presence of a carboxylic acid activating compound  
15 for an aprotic base.

16 In accordance with a still further feature of the invention,  
17 there is provided an assembled hematin, which includes alternating  
18 layers of hematin and a polyelectrolyte on an electrically charged  
19 substrate. Preferably, the polyelectrolyte is cationic.

20 In another embodiment, the invention includes a method of  
21 forming assembled hematin, by alternately depositing one or more  
22 layers of hematin and one or more layers of a polyelectrolyte on  
23 an electrically charged substrate.

24 In accordance with another feature of the invention, there is  
25 provided a method of polymerizing aromatic monomers, such as

1 anilines or phenols. In a preferred embodiment, the  
2 polymerization takes place in the presence of a template.  
3 Typically, the template is anionic.

4 In accordance with a still further feature of the invention,  
5 there is provided a method of polymerizing aromatic monomers by  
6 contacting an aromatic monomer and a template with an assembled  
7 hematin. Preferably, the aromatic monomer is an aniline or a  
8 phenol.

9 In accordance with still another feature of the invention,  
10 there is provided a method of polymerizing an aromatic monomer,  
11 which includes combining the aromatic monomer with a derivatized  
12 hematin catalyst. In a preferred embodiment, the hematin is  
13 derivatized with polyethylene glycoat (PEG). In another preferred  
14 embodiment, the derivatized hematin catalyst and the aromatic  
15 monomer are additionally combined with a peroxide to initiate the  
16 reaction.

17 Advantages of the present invention include resolving the  
18 current limitations of catalysts used in the commercial synthesis  
19 of polyaromatic polymers, by reducing the cost of catalyst and by  
20 providing a catalyst that is active and stable over a wide range  
21 of pHs. The derivatized hematins of the present invention are  
22 water-soluble and recyclable, virtually eliminating the need for  
23 toxic reagents and solvents, and thus creating an environmentally  
24 friendly synthesis for polyaromatic polymers. In addition, the  
25 derivatized hematins of the present invention, in combination with

1 a template, reduce the amount of branching during polymerization,  
2 leading to structurally more consistent product.

3 The present invention is further directed to a syn-enzymatic  
4 polymerization process of PYR and/or EDOT in the presence of SPS,  
5 which results in a novel complex of PPYR and/or PEDOT with SPS,  
6 which has exceptional stability, and good processability.

7 There have been attempts to use different forms of hematin  
8 for catalysis, but it was seen that the catalytic activity was  
9 incomparably lower than that of the enzyme. It is known to  
10 provide for the efficient synthesis of polyaromatic compounds  
11 catalyzed by hematin in mixed solvent systems or buffer systems of  
12 high pH values. It has been found suitable to use a chemically  
13 modified hematin to effectively synthesize conducting polyaniline  
14 in the presence of polyelectrolyte templates. Work in this area  
15 has attempted to manipulate this artificial catalyst towards the  
16 synthesis of conducting PEDOT or PPYR, with the ultimate goal of  
17 expanding the versatility of this hydroxy ferriprotoporphyrin  
18 based catalyst. The method described herein enables the synthesis  
19 of such electroactive polymers, suitable for conductive  
20 transparent coatings.

21 In accordance with a still further feature of the invention,  
22 there is provided a unique template assisted approach for the  
23 synthesis of water-soluble polymers, involving enzymatic  
24 polymerization of aniline and phenol with HRP as the catalyst in  
25 the presence of an anionic polyelectrolyte. In this case, the



1 polyelectrolyte, such as SPS, serves three main functions, namely,  
2 to electrostatically align the aniline monomers to promote a para  
3 directed approach, to provide counterions for doping the polymer,  
4 and to maintain water solubility. Aside from the polyelectrolyte  
5 macromolecular templates, micellar templates like sodium  
6 dodecylbenzene sulphonic acid, and biological templates, like DNA,  
7 have been investigated and seen to be successful nano-reactors in  
8 the one-pot enzymatic synthesis of conducting polyanilines. Thus,  
9 the template provided an environment wherein the pH and the charge  
10 density near the template molecule were different from those of  
11 the bulk solution, the polymerization being carried out at pH 4.0,  
12 (peroxidases are active in the pH range of 4.0 - 8.0).

13 In accordance with still another feature of the invention,  
14 there is provided a novel synthesis of water soluble PEDOT and  
15 PPYR using polyethylene glycoated (PEG) hematin as an efficient  
16 catalyst in the presence of SPS as a template. EDOT and PYR have  
17 been copolymerized using this unique catalyst.

18 The above and other features of the invention, including  
19 various novel details of construction and combinations of steps,  
20 will now be more particularly described with reference to the  
21 accompanying drawings and pointed out in the claims. It will be  
22 understood that the particular methods embodying the invention are  
23 described by way of illustration only and not as limitations of  
24 the invention. The principles and features of this invention may

1 be employed in various and numerous embodiments without departing  
2 from the scope of the invention.

#### 3 4 BRIEF DESCRIPTION OF THE DRAWINGS

5 Reference is made to the accompanying drawings in which are  
6 shown illustrative embodiments of the invention, from which its  
7 novel features and advantages will be apparent.

8 In the drawings:

9 FIG. 1 shows the functionalization of hematin with  
10 polyethylene glycol (PEG) in the presence of N,N'-carbonyl  
11 diimidazole, 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU) and  
12 dimethylformamide (DMF);

13 FIG. 2 shows the Fourier Transform Infrared (FTIR) spectra of  
14 hematin and PEG-hematin. The inset shows an expanded region  
15 between 1500 and 1700cm<sup>-1</sup>;

16 FIG. 3a shows the <sup>1</sup>H NMR spectra of hematin and PEG-hematin  
17 in DMF-d<sub>7</sub>. The inset shows the disappearance of the hematin  
18 carboxylic acid peak when it is derivatized with PEG;

19 FIG. 3b shows the <sup>1</sup>H NMR spectra of hematin and PEG-hematin  
20 in D<sub>2</sub>O;

21 FIG. 4 shows the catalytic activity of hematin and PEG-  
22 hematin for the oxidation of pyrogallol at pH 4.0;

23 FIG. 5 shows the UV-vis absorption spectrum of aniline  
24 monomers and of polyaniline formed during PEG-hematin catalyzed  
25 polymerization;

1           FIG. 6 shows the time dependent UV-vis absorption spectra of  
2           the polyaniline-sodium polystyrene sulfonate (SPS) complex formed  
3           at pH 4 over 2 hours after initiation of polymerization;

4           FIG. 7 shows the pH-dependent UV-vis absorption spectra of  
5           the polyaniline-SPS complex formed after initiation of  
6           polymerization;

7           FIG. 8 shows the UV-vis absorption spectra of polyaniline-SPS  
8           complex as it is titrated with 1 N NaOH and 1 N HCl, demonstrating  
9           that the complex can be reversibly depoded and redoped using base  
10          or acid, respectively;

11          FIG. 9 shows a cycle voltammogram of a solution cast film of  
12          polyaniline-SPS complex synthesized at pH 1.0;

13          FIG. 10 shows the pH-dependent UV-vis absorption spectra of  
14          polyaniline-lignin sulfonate complexes formed during  
15          polymerization;

16          FIG. 11 shows UV-vis absorption spectra of polyaniline-DNA  
17          formed during PEG-hematin catalyzed polymerization;

18          FIG. 12 shows CD spectra of polyaniline-DNA formed during  
19          PEG-hematin catalyzed polymerization;

20          FIG. 13 shows time-dependent UV-vis absorption spectra of the  
21          polymerization of 2-methoxy-5-methylaniline catalyzed by PEG-  
22          hematin;

23          FIG. 14 shows pH-dependent UV-vis absorption spectra of  
24          polyaniline-dodecylbenzenesulfonic acid complexes formed during  
25          polymerization;

1           FIG. 15 shows UV-vis absorption spectra of a SPS-polyphenol  
2           complex formed during polymerization;

3           FIG. 16 is an ultra-violet spectrum of PEDOT templated on SPS  
4           at pH 1.0, 11.0 synthesized using PEG-Hematin;

5           FIG. 17 is an FTIR spectrum of EDOT and SPS-PEDOT collected  
6           after polymerization;

7           FIG. 18 shows an ultra-violet spectra of PYR and PPYR  
8           templated on SPS at pH 2.0, after polymerization thereof;

9           FIG. 19 shows an FTIR spectra of the polymers PYR and PPYR;  
10          and

11          FIG. 20 shows ultra violet spectra of the copolymer of PEDOT  
12          and PPYR templated on SPS, and of the native PYR.

#### 14                           DESCRIPTION OF THE PREFERRED EMBODIMENTS

15          The present invention generally includes a derivatized  
16          hematin and an assembled hematin, along with methods of preparing  
17          the hematins. The invention also includes methods of polymerizing  
18          aromatic monomers in a reaction catalyzed by an assembled hematin  
19          or a derivatized hematin.

20          The methods of the present invention include the use of  
21          hematin, a hydroxyferriprotoporphyrin, which has been derivatized  
22          with one or more non-proteinaceous amphipathic groups. Examples  
23          of amphipathic groups include phosphoglycerides, sphingomyelin,  
24          glycolipids, substituted or unsubstituted polyethers and  
25          polyalkylene glycols, substituted or unsubstituted polyamines such

1 as polyethyleneimine, polyallylamine, and poly(diallylamine);  
2 polyammonium groups, such as poly(allylammonium salts),  
3 poly(trimethylallylammonium salts), poly(triethyallylammonium  
4 salts), poly(dimethyldiallylammonium salts),  
5 poly(diethyldiallylammonium salts), and polysaccharides such as  
6 hydroxypropyl cellulose, hydroxymethyl cellulose, and hydroxyethyl  
7 cellulose.

8 Preferred amphipathic groups include polyalkylene glycols,  
9 such as polyethylene glycol and polypropylene glycol. Preferably,  
10 polyethylene glycol groups have a molecular weight of about 400 to  
11 about 100,000, or more, and preferably a molecular weight of about  
12 5,000 to about 15,000.

13 Aromatic monomers include substituted and unsubstituted  
14 aromatic compounds. Suitable aromatic compounds include 4-(p-  
15 hydroxyphenylazo)pyridine and 4-(p-hydroxyphenylazo)pyridinium  
16 methiodide. Preferred aromatic compounds for polymerization  
17 include aniline, phenol, and 2-methoxy-5-methylaniline.

18 Suitable substituents on aromatic monomers will not  
19 significantly reduce the rate of polymerization as compared to an  
20 unsubstituted aromatic monomer (e.g., will not reduce the rate of  
21 polymerization by more than ten-fold). Examples of suitable  
22 substituents for aromatic monomers include, for example, halogen  
23 (-Br, -Cl, -I, and -F), -OR, -CN, -NO<sub>2</sub>, -COOR, -CONRR<sub>1</sub>, -SO<sub>k</sub>R  
24 (where k is 0, 1, or 2), -NRR<sub>1</sub>, -SR, haloalkyl groups, and -NH-  
25 C(=NH)-NH<sub>2</sub>. R and R<sub>1</sub> are independently, -H, an aliphatic group,

1 and aralkyl group, a heteroaralkyl group, and aromatic group, or a  
2 substituted aromatic group. A substituted aromatic monomer can  
3 have more than one substituent.

4 In a preferred embodiment of the present invention, a  
5 template is combined with the derivatized hematin, an aromatic  
6 monomer, and a peroxide, such that the aromatic monomer aligns  
7 along the template and polymerizes to form a complex including the  
8 polymerized aromatic monomer and the template. A "template," as  
9 that term is employed herein, refers to a polymer or oligomer that  
10 can bind, such as ionically bind, to the aromatic monomer being  
11 polymerized.

12 Suitable template polymers include polyelectrolytes, such as  
13 an anionic polymer or a cationic polymer. Anionic polymer  
14 templates include polymers that include pendant acid functional  
15 groups such as poly(vinylbenzoic acid) and salts thereof,  
16 poly(vinyl polyphosphonic acid) and salts thereof, poly(glutamic  
17 acid) and salts thereof, poly(aspartic acid) and salts thereof,  
18 poly(acrylic acid), and poly(maleic acid co-olefin) and salts  
19 thereof. Co-olefins that can be polymerized with maleic acid to  
20 form poly(maleic acid co-olefin) include 1-propene, 1-butene, 1-  
21 pentene, 1-hexene, 1-heptene, 1-octene, 1-nonene, and 1-decene.  
22 Preferred anionic polymer templates include poly(styrene sulfonic  
23 acid) and salts thereof, lignin sulfonic acid and salts thereof,  
24 and dodecylbenzene sulfonic acid and salts thereof.

1           Optically active templates can be employed in the  
2           polymerization method of the invention. When an optically active  
3           template is employed, the template can induce macro-asymmetry in  
4           the polymerized aromatic monomer due to the close association of  
5           the template with the polymerized aromatic monomer in the complex.  
6           Examples of optically active templates include polynucleic acids  
7           and salts thereof, such as rubonucleic acids and 2'-  
8           deoxyribonucleic acids. Other suitable templates include  
9           biological receptors, peptides, proteins, zeolites, caged  
10          compounds, phenol red, azo compounds, azo polymers, and  
11          dendrimers.

12          In a preferred embodiment, the complex of a polymerized  
13          aromatic monomer and a template is a water-soluble complex of a  
14          polyaniline and a template. Preferably, the polyaniline (pani) is  
15          of the electrically-conducting emeraldine salt form. Emeraldine  
16          is an electrically-conducting form of pani, and has a  
17          characteristic green color when protonated, or doped.

18          In another preferred embodiment, the complex including a  
19          polymerized aromatic monomer and a template is a water-soluble  
20          complex of a polyphenol and a template.

21          In still another preferred embodiment, a polymerized aromatic  
22          monomer complexed to an optically active template has a macro-  
23          asymmetry.

24          A complex of a polymerized aromatic monomer and a template is  
25          prepared by contacting an aromatic monomer, such as an aniline or

1 a phenol, and a template with a derivatized hematin in a solution  
2 of a pH from about 0 to about 12. Preferably, the solution is  
3 buffered, and the pH ranges from about 0 to about 7, and more  
4 preferably ranges from about pH 0 to about pH 4. The ratio of  
5 aromatic monomer to template (measured as the concentration of  
6 template repeat units) can vary from 5:1 to 1:5 (aromatic monomer  
7 : template repeat unit), and is preferably from about 2:1 to about  
8 1:2, and is even more preferably about 1:1.

9 A catalytic amount of the derivatized hematin can be added to  
10 the reaction mixture either before or after addition of the  
11 aromatic monomer. A catalytic amount of the derivatized hematin  
12 is typically between about one unit/mL and five units/mL, where  
13 one unit will form 1.0 mg purpurogallin from pyrogallol in 20  
14 seconds at pH6.0 at 20°C. Preferably, the derivatized hematin is  
15 added to the solution after addition of the template and aromatic  
16 monomer.

17 In a preferred embodiment, a peroxide is also added to the  
18 reaction mixture. The peroxide is added incrementally, such as  
19 not to de-activate the derivatized hematin catalyst, until and  
20 amount approximately stoichiometric with the amount of aromatic  
21 monomer has been added. The reaction can be monitored  
22 spectroscopically.

23 The above polymerization can be carried out in polar solvents  
24 such as ethanol, methanol, isopropanol, dimethylformamide,



1 dioxane, acetonitrile, and diethyl ether, but is preferably  
2 carried out in water.

3 In one embodiment, the present invention is a method of  
4 derivatizing hematin, which includes reacting hematin with one or  
5 more amphipathic compounds, thereby forming a derivatized hematin.  
6 In a preferred embodiment, the hematin is reacted with one or more  
7 amphipathic compounds in the presence of a carboxylic acid  
8 activating compound and an aprotic base. In a more preferable  
9 embodiment, the carboxylic acid activating compound is a  
10 dialkylcarbodiimide. In another preferred embodiment, the  
11 amphipathic compound is a substituted or unsubstituted  
12 polyalkylene glycol. Even more preferably, the polyalkylene  
13 glycol is polyethylene glycol.

14 "Carboxylic acid activating compounds," as used in the  
15 present description, are compounds that serve to couple a  
16 nucleophile, such as a hydroxyl, amine, or thiol group, to a  
17 carboxylic acid, thereby forming an ester, an amide, or a  
18 thioester linkage. Suitable carboxylic acid activating compounds  
19 include dialkylcarbodiimides, preferably diisopropylcarbodiimide  
20 and dicyclohexylcarbodiimide; N,N'-carbonyldiimidazole;  
21 nitrophenol, preferably o-nitrophenol and p-nitrophenol;  
22 pentahalophenol, preferably pentachlorophenol, and  
23 pentabromophenol; N-hydroxysuccinimide; tosyl chloride; 1-  
24 hydroxybenzotriazole; and N-ethyl-N'-(3-dimethylaminopropyl)  
25 carbodiimide.

1            "Aprotic bases," as used herein, include bases without an  
2            exchangeable proton. Suitable aprotic bases include  
3            trialkylamines, such as trimethylamine, triethylamine,  
4            diisopropylethylamine and triphenylamine; pyridine; pyrimidine;  
5            1,8-diazabicyclo[5.4.0]undec-7-3n3 (DBU); and 1,3,5-triazine.

6            Derivatized hematins of the present invention can be  
7            prepared, for example, by reacting about one-half to about ten  
8            mole equivalents of an amphipathic compound, such as polyethylene  
9            glycol, with hematin in the presence of an excess of a carboxylic  
10           acid activating compound, and an aprotic base, in an aprotic  
11           solvent such as dimethylformamide or an ether. The mixture is  
12           allowed to stir for about 6 hours to about 6 days, and is then  
13           quenched with a large volume of water or other protic solvent.  
14           The unreacted reagents are removed by extraction of the reaction  
15           mixture with an organic solvent such as ethyl acetate. The water  
16           layer is concentrated, preferably by lyophilization, to yield the  
17           derivatized hematin.

18           In another embodiment, the present invention is assembled  
19           hematin, which includes one or more layers of hematin alternating  
20           with one or more layers of a polyelectrolyte deposited on a  
21           substrate. In a preferred embodiment, polyelectrolyte is a  
22           cationic polymer, such as a poly(dialkyldiallylammonium salt) or a  
23           poly(trialkylallylammonium salt). Preferably, the polyelectrolyte  
24           is poly(dimethyldiallylammonium chloride).

1           In another embodiment, the present invention includes a  
2       method of polymerizing an aromatic monomer to form a complex of a  
3       polymerized aromatic monomer and a template, by contacting the  
4       aromatic monomer and the template with the assembled hematin.  
5       Preferably, the template is an anionic polymer, such as  
6       poly(styrene sulfonic acid) or a salt thereof. In another  
7       preferred embodiment, the aromatic monomer is a substituted or  
8       unsubstituted aromatic compound, such as an aniline or a phenol.  
9       In yet another preferred embodiment, the complex of the  
10      polymerized aromatic monomer and the template forms in solution or  
11      the complex forms on the assembled hematin. The complex forming  
12      on the assembled hematin can contact one or more layers of hematin  
13      or the polyelectrolyte.

14           In another embodiment, the present invention includes a  
15      method of forming assembled hematin, by alternately depositing  
16      layers of hematin and a polyelectrolyte onto an electrically  
17      charged substrate. Preferably, the polyelectrolyte is a cationic  
18      polymer, and more preferably is a poly(dialkyldiallylammonium  
19      salt) or a (trialkylallylammonium salt, such as poly  
20      (dimethyldiallylammonium chloride).

21           Assembled hematins of the present invention can be prepared,  
22      for example, by dipping a charged substrate, such as a negatively-  
23      charged hydrophilized glass slide, into about 0.1 mM to about 100  
24      mM hematin having a pH from about 6 to about 12 at about 0°C to  
25      about 50°C for about 1 minute to about 100 minutes. The substrate

1 is washed with deionized water and dried with a stream of gas,  
2 such as nitrogen or argon. The substrate with a single layer of  
3 hematin is dipped into about 0.1 mM to about 100 mM  
4 polyelectrolyte having a pH from about 6 to about 12 at about 0°C  
5 to about 50°C for about 1 minute to about 100 minutes. The  
6 substrate is washed with deionized water and dried from a stream  
7 of gas, such as nitrogen or argon. The process can then be  
8 repeated, from about 1 to about 100 times, to produce multiple  
9 alternating layers (or bilayers) of hematin and the  
10 polyelectrolyte on the substrate. For a positively-charged  
11 substrate, the order of dipping into hematin and a polyelectrolyte  
12 is reversed.

13 Polymerizations catalyzed by assembled hematins of the  
14 present invention can be carried out, for example, in a buffered  
15 solution, ranging from about pH 1 to about pH 12, at about 0°C to  
16 about 50°C. An aromatic monomer and a template are added to the  
17 buffered solution, such that the ratio of aromatic monomer to  
18 template repeat unit is about 5 to 1 to about 1 to 5. The  
19 concentration of aromatic monomer is about 0.01 M to about 1 M. A  
20 quantity of assembled hematin, including about 2 to about 100  
21 bilayers of hematin and polyelectrolyte, is added to the solution.  
22 A solution of a peroxide, in an amount sufficient to polymerize  
23 the aromatic monomer, is added dropwise over about 5 minutes to  
24 about 200 minutes. The reaction is maintained for about 1 hour to

1       about 200 hours. The progress of the reaction can be monitored  
2       spectrophotometrically.

3             A peroxide, as used in the present invention, is an organic  
4       or inorganic compound that includes a -O-O- bond, such as ROOR,  
5       where R is as defined above. Preferably, one R is hydrogen, to  
6       give ROOH. Even more preferably, the peroxide is hydrogen  
7       peroxide, HOOH.

8             Suitable substrates for assembled hematin are any solids that  
9       can maintain an electrical charge. Examples of substrates include  
10      glasses (e.g., pyrex and glass slides), plastics (e.g., poly(vinyl  
11      chloride) and poly(ethylene)), ceramics, metals, and the like.  
12      Preferred substrates are glass slides, which have been  
13      hydrophilized with an aqueous alkali solution, such as Chem-solv,  
14      under ultrasonication.

15            The functionalities of the polymers may be tuned to impart  
16      requisites, such as sensing, electrochemical, optical and  
17      electronic properties through copolymerization with functionalized  
18      monomers. The polymers have sites for further modifications, such  
19      as covalently coupling other functionalities and even biomolecules  
20      through simple coupling chemistry.

21            The conducting polymers in these polymer complexes will allow  
22      for use in a wide range of applications including, but not limited  
23      to, chemical and biological sensing, electrostatic shielding,  
24      photovoltaic cell corrosion protection, light rechargeable  
25      batteries, flexible light-emitting diodes, electrochromic devices,

1 smart windows, chaff materials, electromagnetic radiation  
2 absorbers and modulators, and drug delivery systems.

3 Accordingly, to achieve the foregoing objects and in  
4 accordance with the purpose of the invention, as embodied and  
5 broadly described herein, a method for matrix assisted, syn-  
6 enzyme-catalyzed polymerization or copolymerization of PYR, PEDOT  
7 and aniline comprises the preparation of an aqueous solution  
8 containing PYR and/or PEDOT, SPS, Hem-PEG syn-enzyme and a  
9 reaction initiator (hydrogen peroxide). The procedure is a one-  
10 step, in situ reaction, which is highly selective and which  
11 produces minimal by-products and chemical waste. The resulting  
12 polymers or copolymers solution can be used immediately as is or  
13 purified via such techniques as dialysis and centrifugation.

14 Matrix materials may include, but are not limited to,  
15 electrolytes which have various aromatic backbones and/or pendant  
16 groups, aliphatic backbones and/or pendant groups, optically  
17 active (chromophoric) backbones and/or pendant groups,  
18 electrically active backbones and/or pendant groups and various  
19 degrees of sulfonation/functionalization. The ionized groups on  
20 these electrolyte matrices may include and are not limited to  
21 sulfonates, carboxylates, phosphates, and borates. Manipulation  
22 of the molecular weight and purity of the matrices will allow for  
23 optimized polymerization and processing conditions.

24 The present invention is premised on the discovery that  
25 unsurpassed electrical and optical stability, processability,

1 tunability and environmental compatibility are imparted to a new  
2 matrix assisted syn-enzymatic polymerization of EDOT, pyrrole,  
3 aniline and phenol. In addition, with judicious choice of matrix  
4 and/or monomer, the final polymer complex properties may be  
5 tailored to suit a wide range of real device applications.

6 The present invention will now be further described by the  
7 following non-limiting examples.

#### 8 9 EXAMPLE 1

##### 10 Synthesis of PEG-Hematin complex

11 The PEG-hematin complex was obtained through the coupling of  
12 PEG chains to a hematin molecule through ester linkages as shown  
13 in FIG. 1. The PEG-hematin complex was prepared by the addition  
14 of a mole equivalent of PEG (19 mg) to hematin (200 mg) in the  
15 presence of activators N, N'-carbonyldiimidazole (0.05 g) and 1,8-  
16 diazabicyclo[5.4.0]undec-7-ene (DBU) (0.047 g) in DMF. The  
17 mixture was allowed to stir for 48 hours then was quenched by the  
18 addition of a large volume of deionized water. The unreacted  
19 reagents were removed by extraction with ethyl acetate. The water  
20 layer was subsequently lyophilized to yield PEG-hematin as a  
21 reddish-brown solid.

22 The complex was characterized using NMR and FTIR  
23 spectroscopy. The average extent of modification of the acidic  
24 groups of hematin was determined using UV-vis spectroscopy. The  
25 UV-vis spectra of the PEG-hematin exhibited a decrease in the

1 Soret band (420 nm), a porphyrin centered  $\pi$ - $\pi^*$  transition, in  
2 comparison to hematin, which was used to calculate the amount of  
3 hematin present in the sample. However, the energy and spectral  
4 bandwidths of PEG-hematin were indistinguishable from hematin,  
5 which indicated that the modification of hematin by poly (ethylene  
6 glycol) does not affect the heme structure. Based on this  
7 assumption, the average concentration of hematin in the PEG-  
8 hematin sample was subsequently determined to be 67% by weight.

9 An FTIR spectrum of PEG-hematin indicated the presence of an  
10 ester functionality by the appearance of a doublet at 1646 and  
11 1651  $\text{cm}^{-1}$  (similar to diethyl phthalate) accompanied by the  
12 complete disappearance of the peak at 1712  $\text{cm}^{-1}$  for the acid  
13 carbonyl of hematin (FIG. 2). The strong peak at 1100  $\text{cm}^{-1}$   
14 corresponded to the ether linkage of the glycol moiety.

15 An  $^1\text{H}$  NMR spectrum of PEG-hematin in  $\text{DMF-d}_7$  shows the  
16 disappearance of the peak at 10.2 ppm, which was assigned to the  
17 carboxylic proton of hematin (FIG. 3a). This clearly indicated  
18 that the carboxylic acid hydroxyl moiety was transformed into an  
19 ester. The large broad peak at 3.8 ppm was assigned to the poly  
20 (ethylene glycol) protons. However, the spectra could not be well  
21 resolved in the region of 2-4 ppm due to the interference of the  
22 peaks assigned to the residual protons in deuterated DMF. In  
23 order to get a better resolution of the spectrum, the solvent  
24 system was changed to deuterated water. The spectrum  $\text{D}_2\text{O}$  could  
25 not be used to distinguish the absence of the carboxylic acid



1 proton due to proton exchange with D<sub>2</sub>O. However, comparison of  
2 the spectrum of PEG-hematin and spectrum of poly(ethylene glycol),  
3 in D<sub>2</sub>O showed the changes in the position of the PEG peaks of PEG-  
4 hematin in comparison to PEG alone. It was found the PEG  
5 exhibited a major peak at 3.8 ppm, which was assigned to the bulk  
6 of the polymer chains, while the adjoining peaks (triplets) were  
7 assigned to the end groups of the polymer. When a PEG-hematin  
8 derivative was formed, the peak at 4.0 ppm shifted upfield and  
9 merged into the main peak. This was accompanied by considerable  
10 broadening and a shift of the peak at 3.8 ppm to 3.6 ppm (FIG.  
11 3b). It was postulated that methylene protons  $\alpha$  to the hydroxy  
12 group PEG, on being attached by an ester linkage to hematin,  
13 shifted upfield while methylene protons  $\beta$  to the hydroxy groups of  
14 PEG were affected by the inhomogeneous paramagnetic environment,  
15 leading to broadening. These observed changes strongly indicated  
16 the formation of an ester bond between PEG and hematin.

17 The activity of the PEG-hematin was assessed through the  
18 oxidation of pyrogallo (0.5%) to purpurogallin in 14 mM potassium  
19 phosphate buffer in the presence of 0.027% (w/w) hydrogen  
20 peroxide. The activity of the PEG-hematin was found to be  
21 approximately 30-fold higher as compared to native hematin at a pH  
22 4.0 (FIG. 4). It is believed that the activity of hematin is  
23 dependent on its solubility. Thus, the enhanced activity of the  
24 PEG-hematin is attributed to its enhanced solubility.

## EXAMPLE 2

### Synthesis of Polyaniline (Pani)

The polymerization of aniline was carried out in 0.1 M sodium phosphate buffer (10 mL) maintained at pH 1. To this buffer solution the aniline monomer was added. The catalyst, PEG-hematin (60  $\mu$ g), was added only just prior to the addition of hydrogen peroxide. The polymerization was initiated by the incremental addition of a stoichiometric amount of hydrogen peroxide, with respect to aniline. 0.3%  $\text{H}_2\text{O}_2$  (w/v) was used with constant stirring and the progress of the reaction was monitored spectroscopically (FIG. 5). Typically, all reaction systems were left stirred until completion of polymerization followed by precipitation of the pani. The pani synthesized was filtered off and thoroughly washed with acetone a few times followed by drying in a vacuum oven. The conductivity of the pani pellet was found to be of the order of 0.2 S/cm.

This reaction thus proved the versatility and ability of the PEG-Hematin for the synthesis of stable conducting pani even in the absence of template. The pani formed in this case was again redox reversible as proved by cyclic voltammetry studies.

## EXAMPLE 3

### Synthesis of Sodium Poly (sodium-4-styrenesulfonate)-Polyaniline Complex

1           The polymerization of aniline was carried out in 0.1 M sodium  
2   phosphate buffer over a range of pH conditions from pH 1-4. A 17  
3   mM solution of SPS template in phosphate buffer (100mM) was  
4   prepared to which the aniline monomer was added in a 1:1 molar  
5   ratio of aniline to sodium styrene sulfonate monomer. The  
6   catalyst, PEG-hematin (5 mg), was added just prior to the addition  
7   of hydrogen peroxide. The polymerization was initiated by the  
8   incremental addition of a stoichiometric amount of hydrogen  
9   peroxide (relative to aniline). In all cases, 0.3% H<sub>2</sub>O<sub>2</sub> (w/v) was  
10   used with constant stirring, and the progress of the reaction was  
11   monitored spectroscopically. On completion of polymerization, the  
12   solution was transferred to individual regenerated natural  
13   cellulose membrane bags (molecular weight cut-off 10,000 D) and  
14   were dialyzed against 5000 mL of acidified deionized water  
15   maintained at pH 4.0 to remove unreacted monomers and oligomers.  
16   The solid SPS-Pani complex was obtained by evaporation of the  
17   deionized water followed by drying in a vacuum oven.

18           It was observed that the solution slowly turned dark green,  
19   indicating the formation of the doped emeraldine salt form of  
20   conducting pani. The UV-vis absorption spectra of the Pani/SPS  
21   complex, formed at different time intervals over a period of 2  
22   hours at pH 4.0 after initiation of polymerization reaction, is  
23   shown in FIG. 6. The UV-vis spectra showed the presence of  
24   polaron absorption bands at 400 nm and 800-1200 nm, which was  
25   consistent with the formation of the conducting form of pani.

1 This polymerization was also carried out at different pH values  
2 ranging from pH 1.0 to pH 4.0 as shown in FIG. 7. The formation  
3 of pani was observed in all cases, thus demonstrating the  
4 stability and robustness of the PEG-hematin in comparison to  
5 hematin (insoluble at low pH) or horseradish peroxidase, HRP  
6 (denatured at low pH). Also, the pani formation reaction  
7 catalyzed by PEG-hematin was found to be complete with greater  
8 than 90% yield within a few hours, while the unmodified hematin  
9 showed little or no reactivity within the same time period under  
10 these acidic conditions.

11 The redox tunability of the pani formed was further  
12 demonstrated by dedoping the emeraldine salt form of pani at high  
13 pH and then redoping with acid. With increasing pH (dedoping) on  
14 titration with 1 N NaOH, the polaron bands at 400 nm and 800 nm  
15 were found to diminish, while a new band at 600 nm began to emerge  
16 due to the exciton transition of the quinoid ring giving rise to a  
17 blue solution indicating that the pani had been fully dedoped to  
18 the base form. On titrating the solution back with 1 N HCl  
19 (redoping), a reversible color change was observed and the spectra  
20 is shown in FIG. 8. Furthermore, an isosbestic point at 710 nm  
21 was also observed, which was indicative of the changes in the pani  
22 oxidation state. This behavior was similar to the pani  
23 synthesized chemically or enzymatically with HRP and confirmed the  
24 formation of the conducting pani emeraldine salt form  
25 (electroactive form) catalyzed by PEG-hematin.

1           The conductivity of the emeraldine salt form of pani  
2 synthesized at pH less than 4 was found to be about  $10^{-3}$  S/cm.

3           Furthermore, cyclic voltammetry studies were carried out to  
4 determine the electrochemical nature of pani synthesized by the  
5 PEG-hematin catalysis. The cyclic voltammogram of a cast film of  
6 an SPS-pani complex (FIG. 9) showed two sets of peaks indicating  
7 two reversible redox cycles at a scan rate of 100 mV/s over a  
8 potential window of -0.2-1.2V.

#### 10       EXAMPLE 4

##### 11       Synthesis of Lignosulfonate-pani Complex

12           5.2 mg of a lignin sulfonate polyelectrolyte complex was  
13 dissolved in 10 mL of sodium monophosphate buffer (0.1 M)  
14 maintained at pH 4.0. This was followed by the addition of 18  $\mu$ L  
15 of aniline, a catalytic amount of PEG-Hematin and an amount of  
16 hydrogen peroxide (0.3%) stoichiometric with aniline. The  
17 reaction mixture was allowed to stir until precipitation of the  
18 polyelectrolyte-pani complex ceased. The reaction was also  
19 carried out in solutions having pHs ranging from pH 1-4 (FIG. 10).  
20 The precipitated lignin sulfonate-pani complex obtained was washed  
21 several times with acidified acetone to remove the unreacted  
22 monomer and finally washed with acidified deionized water,  
23 filtered under suction through a polycarbonate filter and dried in  
24 a vacuum oven to yield lignin sulfonate pani complex.

1           When the polymerization was conducted at pH 3.0, there was a  
2 peak of low intensity at 767 nm for the emeraldine form of pani,  
3 which was completely absent during polymerization at pH 4.0. The  
4 extended absorption of 1200 nm indicated the formation of the  
5 extended conjugation of the pani backbone. Thus, the synthesis of  
6 pani complexed with a natural polymer further widens the scope of  
7 applications to other natural polyelectrolytes to form versatile,  
8 environmentally benign conducting polymers.

#### 10       EXAMPLE 5

##### 11       Synthesis of DNA-pani Complex

12           The polymerization of aniline in the presence of Calf Thymus  
13 DNA was carried out in sterile 10 mM phosphate buffer. A 1.0 mM  
14 calf thymus DNA solution was prepared by dissolving the required  
15 amount of DNA in 10 mL of sterilized sodium phosphate buffer  
16 maintained at pH 4. The concentration of DNA was determined by  
17 the UV absorbance at 258 nm. To this DNA solution, 4.5  $\mu$ l (5 mM)  
18 of aniline was added. The pH of the solution was again checked  
19 and adjusted to 4.3, and 5 mg of PEG-Hematin were added. To this  
20 reaction mixture, a solution of hydrogen peroxide (0.3% solution,  
21 4.5  $\mu$ l, 5 mM) was added drop-wise, to initiate the polymerization  
22 and reaction of aniline was followed using UV-Vis spectroscopy and  
23 circular dichroism polarimetry.

24           When the aniline monomer was added to a DNA solution at pH  
25 4.3, the electrostatic interaction between the protonated aniline

1 monomers and the phosphate groups in the DNA caused the monomers  
2 to closely associate with the DNA. The association of the  
3 protonated aniline monomer on the DNA template facilitated a  
4 predominantly *para*-directed coupling and inhibited parasitic  
5 branching during the polymerization. The high proton  
6 concentration around the phosphate groups also provided a unique  
7 local lower pH environment that permitted the polymerization of  
8 aniline at a higher pH than that necessary with conventional  
9 chemical polymerization of aniline. The polymerization was  
10 catalyzed by PEG-hematin and initiated by hydrogen peroxide.  
11 However, as the polymerization proceeded over a period of time and  
12 a critical chain length was attained, the DNA-pani complex  
13 precipitated out of solution.

14 It was concluded that the complex remained soluble as long as  
15 there were enough phosphate groups on the DNA available for  
16 solvation. As the polymerization proceeded, the preferred  
17 molecular interaction between the charged aniline groups and the  
18 phosphate groups of DNA caused the growing chain to occupy a  
19 majority of these sites leading to the salting out of the DNA-pani  
20 complex. The polymerization reaction was followed using UV-vis  
21 spectroscopy and circular dichroism polarimetry.

22 The UV-vis spectra of the DNA-pani complex recorded after  
23 initiation of the polymerization are shown in FIG 11. The UV-vis  
24 absorbance spectra showed a peak around 260 nm emerging from the  
25 absorption of the base pairs of DNA along with polaron absorption

1 bands at 420 nm and 750 nm, indicating the formation of the  
2 conducting emeraldine salt form of pani.

3 The bases of the nucleic acid have a plane of symmetry and  
4 thus are not intrinsically optically active. However, the  
5 deoxyribose sugar is asymmetric and since the bases are attached  
6 to the anomeric carbon of these sugars, the sugar can induce a  
7 circular dichroism in the absorption bands of the bases. These  
8 bands may be observed either for the intensely electronically  
9 allowed  $\pi$ - $\pi^*$  transitions, or for the weakly allowed  $n$ - $\pi^*$   
10 transitions because these transitions are magnetically allowed.  
11 Also, the  $\pi$  electron systems of the bases make them hydrophobic,  
12 so the bases tend to stack in hydrogen-bonding solvents to  
13 minimize the  $\pi$ -electron surface area exposed to the solvent. The  
14 hydrophobic planes and hydrophilic edges as well as charge-charge  
15 interactions cause the bases to stack and the polymer to adopt a  
16 helical structure.

17 Preferential handedness is induced in these helical  
18 structures by the intrinsically asymmetric sugars, giving the DNA  
19 polymer a whole super asymmetry. The electronic transitions of  
20 these chromophoric bases are in close proximity and can thus  
21 interact to give well-defined CD spectra. The CD spectrum of the  
22 DNA-pani complex showed a reduction in the intensity of the peak  
23 at 275 nm (FIG. 12).

24 This change indicated a polymorphic transition in DNA causing  
25 the DNA to change from a loosely wound form to the over-wound



1 form. The appearance of a positive peak at 450 nm indicated that  
2 the helical polyelectrolyte DNA template induces a macroscopic  
3 order in the pani that is formed. This result proves the  
4 extensive versatility of the PEG-Hematin catalyst with a variety  
5 of templates, including delicate biomacromolecules, in providing  
6 the optimal catalytic activity for polymerization.

#### 8 EXAMPLE 6

##### 9 Synthesis of Poly(2-methoxy-5-methylaniline)-SPS complex

10 The polymerization of 2-methoxy-5-methylaniline (2M5M) was  
11 carried out in 0.1 M sodium phosphate buffer of pH 4.0. A 17 mM  
12 SPS template solution, as measured from the concentration of  
13 sodium styrene sulfonate monomers, in phosphate buffer (10 mL) was  
14 prepared, to which 2M5M (24 mg) was added in the desired (1:1,  
15 2M5M:SPS) molar ratio. The polymerization was initiated after  
16 addition of 5 mg of PEG-Hematin, by the incremental addition of an  
17 amount of peroxide (0.3% w/v) stoichiometric with 2M5M, with  
18 constant stirring. The progress of the reaction was monitored  
19 spectroscopically. After the reaction was complete, the solution  
20 was dialyzed to remove the unreacted monomers, followed by  
21 evaporation to yield a SPS-poly(2M5M) complex.

22 The UV-vis absorption spectra of the poly(2M5M)/SPS complex  
23 formed is shown in FIG. 14. The spectra again showed the presence  
24 of a polaron band at 425 nm and extended conjugation in the longer  
25 wavelength range indicating the linear conducting form of pani.

1 This polymer also showed reversible redox tunability similar to  
2 that observed for the SPS-Pani complex formed in Example 2. The  
3 SPS-poly(2M5M) formed could also be reversibly de-doped on  
4 titrating with 1N NaOH and re-doped by back titrating with 1N HCL.

#### 6 EXAMPLE 7

##### 7 Synthesis of Sodium Dodecylbenzenesulfonic Acid-Pani Complex

8 Polymerization of aniline was carried out in 0.1 M sodium at  
9 pH 4. A 17 mM solution of dodecylbenzenesulfonic acid (DBSA) in  
10 phosphate buffer (100 mM) was prepared to which the aniline  
11 monomer was added in the desired (1:1, Aniline:DBSA) molar ratio.  
12 The catalyst, PEG-Hematin (5 mg), was added just prior to the  
13 addition of hydrogen peroxide. The polymerization was initiated  
14 by the incremental addition of an amount of hydrogen peroxide  
15 stoichiometric to aniline. In all cases, 0.3% H<sub>2</sub>O<sub>2</sub> (w/v) was used  
16 with constant stirring. The progress of the reaction was  
17 monitored spectroscopically.

#### 19 EXAMPLE 8

##### 20 Synthesis of SPS-Polyphenol Complex

21 A polymerization reaction was carried out in 10 mL of aqueous  
22 phosphate buffer (100 mM). The pH of the reaction media for the  
23 phenol polymerization was maintained at pH 7.0 and equimolar  
24 concentrations (17 mM) of SPS, with respect to the concentration  
25 of the repeat units, and phenol monomer were added to the buffered

1 solution, followed by 10 mg of the PEG-hematin. The reaction was  
2 initiated by addition of a stoichiometric, with respect to phenol,  
3 amount  $\text{H}_2\text{O}_2$  (30% w/v) in one lot to facilitate the formation of  
4 high molecular weight polypenol. The reaction was monitored  
5 spectroscopically. A control experiment was also carried out  
6 simultaneously in the absence of catalyst. The final products  
7 were dialyzed using Centricon concentrators (10,000 Mw cut off,  
8 Amicon Inc., Beverly, MA) to remove unreacted monomers. The  
9 samples were then dried under vacuum at 50°C and used for further  
10 analysis. The yield was calculated to be typically 95% or higher.

11 The PEG-hematin complex was also found to catalyze the  
12 polymerization of phenol at pH 7.0 more efficiently than that  
13 compared to the native hematin and peroxidase (FIG. 15). The  
14 large broad absorption tail in the region from 300-700 nm  
15 conferred the presence of extended conjugation and indicated  
16 formation of polyphenol by PEG-hematin reaction. In comparison,  
17 the absorption of the hematin-catalyzed reaction was relatively  
18 weak. Thus, modification of the hematin with PEG was observed to  
19 significantly improve the reactivity to suit the desired reaction  
20 conditions leading to the formation of polyphenol.

## 21 22 EXAMPLE 9

### 23 Preparation of Assembled Hematin

24 Glass slides (25 by 75 mm) were hydrophilized with 1% Chem-  
25 solv solution in deionized water under ultrasonication for use as

1 substrates. This treatment generates negative charges on the  
2 surface of the slides due to partial hydrolysis. After 3 hours,  
3 the slides were ultrasonicated twice in deionized water for 30  
4 minutes before use.

5 The electrostatic layer-by-layer deposition process was  
6 carried out in two steps. Poly (diallyldimethylammonium chloride)  
7 (PDAC) (10 mM) and hematin (3 mM) solutions were prepared over a  
8 pH range from 5 to 11. In the first step, hydrophilized glass  
9 slides were immersed in PDAC solution for 10 minute at room  
10 temperature and washed with deionized water for 5 minutes. After  
11 the deposition and washing steps, the slides were dried with a  
12 stream of nitrogen. In the second step, the substrates with a  
13 single layer of PDAC were immersed into the hematin solution for  
14 10 minutes and subsequently washed with deionized water and dried  
15 with a stream of nitrogen to produce an assembled hematin, having  
16 a bilayer film of PDAC/hematin. This dipping procedure was  
17 iterated to build up multilayer films.

#### 18 19 EXAMPLE 10

#### 20 Synthesis of Pani-SPS Complex Using Assembled Hematin

21 Polymerization of aniline was carried out at room temperature  
22 in a 40mL, 0.1M phosphoric acid buffer solution, which contained a  
23 1:1 molar ratio of SPS (MW 1,000,000; moles correspond to quantity  
24 of monomers units) to aniline 0.167g (0.81mmol). SPS was added  
25 first to the buffered solution, followed by an addition of 2.1mL

1 of aniline stock solution (0.036mL aniline to 1 mL buffer at pH  
2 1.4) with constant stirring. A seventeen bilayer Hematin/PDAC  
3 assembly was immersed in the solution. To initiate aniline  
4 polymerization, 11mL of 0.25% H<sub>2</sub>O<sub>2</sub> was added dropwise,  
5 incrementally, over 30 minutes. The reaction was maintained for  
6 24 hours, and carried out at different pH values (1.0, 2.0, 3.0).  
7 The rate of assembled hematin catalyzed polymerization was  
8 monitored by a Perkin-Elmer Lambda-9-UV-vis spectrophotometer at  
9 room temperature.

#### 11 EXAMPLE 11

#### 12 Synthesis of Pyrrole

13 Pyrrole polymerization in presence of SPS was catalyzed by  
14 PEG-Hematin at 25°C using hydrogen peroxide under ambient  
15 conditions. 36.8 mg of SPS was dissolved in deionized water (10  
16 ml) at pH 2.0 for the polymerization of pyrrole (0.2 mM). This  
17 was followed by the addition of 5 mg of PEG-Hematin to this  
18 solution. The polymerization was initiated by the addition of  
19 several aliquots of 800µl of 0.03% hydrogen peroxide added in  
20 several small increments. The reactants were stirred for 12 hours  
21 to complete the polymerization followed by dialysis using  
22 Centricon concentrators. The samples were then dried under vacuum  
23 at 60°C and used for further analysis. The gravimetric yield was  
24 typically 95% higher.

1       EXAMPLE 12

2       Synthesis of Poly (3,4)-ethylenedioxythiophene (PEDOT)

3           PEDOT polymerization in presence of SPS was catalyzed by PEG-  
4       Hematin at 25°C using hydrogen peroxide under ambient conditions.  
5       36.8 mg of SPS was dissolved in deionized water (10 ml) at pH 1.0  
6       for the polymerization of PEDOT (concentration 0.2 mM). This was  
7       followed by the addition of 5 mg of PEG-Hematin to this solution.  
8       The polymerization was initiated by the addition of several  
9       aliquots of 800 µl of 0.03% hydrogen peroxide added in several  
10      small increments. The reactants were stirred for 12 hours to  
11      complete the polymerization followed by dialysis using Centricon  
12      concentrators (10,000 cut off). The samples were then dried under  
13      vacuum at 60°C and used for further analysis. The gravimetric  
14      yield was typically 95% or higher.

15  
16      EXAMPLE 13

17      Synthesis of copolomers using Pyrrole and (3,4)-  
18      Ethylenedioxythiophene (EDOT) as monomers

19           Pyrrole (concentration 0.2 mM) and EDOT (0.2 mM)  
20      copolymerization in presence of SPS was catalyzed by PEG-Hematin  
21      at 25°C using hydrogen peroxide under ambient conditions. 36.8 mg  
22      of SPS was dissolved in deionized water (10 ml) at pH 1.0. This  
23      was followed by the addition of 5 mg of PEG-Hematin to this  
24      solution. The copolymerization was initiated by the addition of

1 several aliquots of 800  $\mu$ l of 0.03 % hydrogen peroxide added in  
2 several small increments. The reactants were stirred for 12 hours  
3 to complete the copolymerization followed by dialysis using  
4 Centricon concentrators. The samples were then dried under vacuum  
5 at 60°C and used for further analysis. The gravimetric yield was  
6 typically 95% or higher.

#### 8 EXAMPLE 14

9 Synthesis of copolymeres using Pyrrole and EDOT as monomers

10 Pyrrole (concentration 0.2 mM) and EDOT (0.2 mM)  
11 copolymerization in the presence of SPS was catalyzed by PEG-  
12 Hematin at 25°C using hydrogen peroxide under ambient conditions.  
13 36.8 mg of SPS was dissolved in deionized water (10 ml) at pH 2.0.  
14 This was followed by the addition of 5 mg of PEG-Hematin to this  
15 solution. The copolymerization was initiated by the addition of  
16 several aliquots of 800 $\mu$ l of 0.03% hydrogen peroxide added in  
17 several small increments. The reactants were stirred for 12 hours  
18 to complete the copolymerization followed by dialysis using  
19 Centricon concentrators. The samples were then dried under vacuum  
20 at 60°C and used for further analysis. The gravimetric yield was  
21 typically 95% or higher.

#### 23 EXAMPLE 15

24 Synthesis of copolymers using Pyrrole and Aniline as monomers

1 Pyrrole (concentration 0.2 mM) and aniline (0.2 mM)  
2 copolymerization in presence of SPS was catalyzed by PEG-Hematin  
3 at 25°C using hydrogen peroxide under ambient conditions. 36.8 mg  
4 of SPS was dissolved in deionized water (10 ml) at pH 2.0. This  
5 was followed by the addition of 5 mg of PEG-Hematin to this  
6 solution. The copolymerization was initiated by the addition of  
7 several aliquots of 800µl of 0.03% hydrogen peroxide added in  
8 several small increments. The reactants were stirred for 12 hours  
9 to complete the polymerization followed by dialysis using  
10 Centricon concentrators. The samples were then dried under vacuum  
11 at 60°C and used for further analysis. The gravimetric yield was  
12 typically 95% or higher.

#### 14 EXAMPLE 16

15 Synthesis of copolymers using Aniline and EDOT as monomers

16 Aniline (concentration 0.2 mM) and EDOT (0.2mM)  
17 copolymerization in presence of SPS was catalyzed by PEG-Hematin  
18 at 25°C using hydrogen peroxide under ambient conditions. 36.8 mg  
19 of SPS was dissolved in deionized water (10 ml) at pH 2.0. This  
20 was followed by the addition of 5 mg of PEG-Hematin to this  
21 solution. The copolymerization was initiated by the addition of  
22 several aliquots of 800µl of 0.03% hydrogen peroxide added in  
23 several small increments. The reactants were stirred for 12 hours  
24 to complete the copolymerization followed by dialysis using



1       Centricon concentrators. The samples were then dried under vacuum  
2       at 60°C and used for further analysis. The gravimetric yield was  
3       typically 95% or higher.

#### 5       EXAMPLE 17

6       Synthesis of copolymers using Pyrrole, Aniline and EDOT as  
7       monomers

8             Pyrrole (concentration 0.2 mM), aniline (0.2mM) and EDOT (0.2  
9       mM) copolymerization in presence of SPS was catalyzed by PEG-  
10       Hematin at 25°C using hydrogen peroxide under ambient conditions.  
11       36.8 mg of SPS was dissolved in deionized water (10 ml) at pH 2.0.  
12       This was followed by the addition of 5 mg of PEG-Hematin to this  
13       solution. The copolymerization was initiated by the addition of  
14       several aliquots of 800µl of 0.03% hydrogen peroxide added in  
15       several small increments. The reactants were stirred for 12 hours  
16       to complete the copolymerization followed by dialysis using  
17       Centricon concentrators. The samples were then dried under vacuum  
18       at 60°C and used for further analysis. The gravimetric yield was  
19       typically 95% or higher.

20  
21       This invention provides a significant advancement over  
22       current methods used for the synthesis of a conducting and  
23       processable form of polypyrrole and PEDOT. This method addresses  
24       and resolves processability and stability problems, which have

1       limited the commercial use of polypyrrole and PEDOT. The syn-  
2       enzymatic synthesis provides a specific, simple and  
3       environmentally friendly synthetic approach, while the SPS  
4       provides stability and processability. In addition, the  
5       SPS/polypyrrole and SPS/PEDOT complex described herein is expected  
6       to transition effectively into may established applications where  
7       conductivity is desirable.

8               While this invention has been particularly shown and  
9       described with references to preferred embodiments thereof, it  
10      will be understood by those skilled in the art that various  
11      changes in form and details may be made therein without departing  
12      from the scope of the invention encompassed by the appended  
13      claims.